

Applicants: Christophe P.G. Gerald, et al.
Serial No.: 09/866,248
Filed: May 25, 2001
Page 3

Amendments to the claims:

This listing of claims will replace all prior versions, and listings, of claims in the application:

Listing of the claims:

--1-182 (Canceled)

--183: (Currently amended) A process for preparing a ~~pharmaceutical~~ composition which comprises: ~~determining whether a compound is a mammalian NPFF receptor agonist using the method of:~~

(a) determining whether a compound is a mammalian NPFF receptor agonist by a method which comprises contacting cells transfected with and expressing DNA encoding the mammalian NPFF receptor with the compound under conditions permitting the activation of the mammalian NPFF receptor, and detecting an increase in mammalian NPFF receptor activity, so as to thereby determine whether the compound is a mammalian NPFF receptor agonist;

(b) recovering the compound free of any mammalian NPFF receptor; and

(c) ~~admixing a pharmaceutically acceptable amount of the compound with a pharmaceutically acceptable carrier, thereby preparing the pharmaceutical composition;~~

wherein the mammalian NPFF receptor comprises an amino

acid sequence which is the same as 1) the sequence of the human NPFF1 receptor encoded by plasmid pWE15-hNPFF1 (ATCC Accession No. 203183) or by plasmid pCDNA3.1-hNPFF1 (ATCC Accession No. 203605); 2) the sequence of the rat NPFF1 receptor encoded by plasmid pEXJ-rNPFF1 (ATCC Accession No. 203184); 3) the sequence of the human NPFF2 receptor encoded by plasmid pCDNA3.1-hNPFF2b (ATCC Accession No. 203255); or 4) the sequence shown in SEQ ID NO: 2, 4, 6 or 8.

--184: (Currently amended) A process for preparing a ~~pharmaceutical~~ composition which comprises: ~~determining whether a compound is a mammalian NPFF receptor agonist using the method of:~~

- (a) determining whether a compound is a mammalian NPFF receptor agonist by a method which comprises contacting cells transfected with and expressing DNA encoding the mammalian NPFF receptor with the compound under conditions permitting the activation of the mammalian NPFF receptor, and detecting an increase in mammalian NPFF receptor activity, so as to thereby determine whether the compound is a mammalian NPFF receptor agonist;
- (b) recovering the compound free of any mammalian NPFF receptor; and
- (c) ~~admixing a pharmaceutically acceptable amount of the compound with a pharmaceutically acceptable carrier,~~ thereby preparing the ~~pharmaceutical~~ composition;

wherein the mammalian NPFF receptor comprises an amino

Applicants: Christophe P.G. Gerald, et al.
Serial No.: 09/866,248
Filed: May 25, 2001
Page 5

acid sequence which is the same as 1) the sequence of the human NPFF1 receptor encoded by plasmid pWE15-hNPFF1 (ATCC Accession No. 203183) or by plasmid pCDNA3.1-hNPFF1 (ATCC Accession No. 203605); 2) the sequence of the rat NPFF1 receptor encoded by plasmid pEXJ-rNPFF1 (ATCC Accession No. 203184); 3) the sequence of the human NPFF2 receptor encoded by plasmid pCDNA3.1-hNPFF2b (ATCC Accession No. 203255); or 4) the sequence shown in SEQ ID NO: 2, 4, 6 or 8.

--185: (Currently amended) A process for preparing a ~~pharmaceutical~~ composition which comprises:

- (a) identifying a chemical compound which specifically binds to a mammalian NPFF receptor by a method which comprises contacting cells containing DNA encoding and expressing on their cell surface the mammalian NPFF receptor, wherein such cells do not normally express the mammalian NPFF receptor, or a membrane preparation of such cells, with the chemical compound under conditions suitable for binding, and detecting specific binding of the chemical compound to the mammalian NPFF receptor;
- (b) recovering the compound free of any mammalian NPFF receptor; and
- (c) admixing a ~~pharmaceutically acceptable amount~~ the compound with a ~~pharmaceutically acceptable~~ carrier, thereby preparing the ~~pharmaceutical~~

composition;

wherein the mammalian NPFF receptor comprises an amino acid sequence which is the same as 1) the sequence of the human NPFF1 receptor encoded by plasmid pWE15-hNPFF1 (ATCC Accession No. 203183) or by plasmid pcDNA3.1-hNPFF1 (ATCC Accession No. 203605); 2) the sequence of the rat NPFF1 receptor encoded by plasmid pEXJ-rNPFF1 (ATCC Accession No. 203184); 3) the sequence of the human NPFF2 receptor encoded by plasmid pCDNA3.1-hNPFF2b (ATCC Accession No. 203255); or 4) the sequence shown in SEQ ID NO: 2, 4, 6 or 8.

--186: (Currently amended) A process for preparing a ~~pharmaceutical~~ composition which comprises:

- (a) identifying a chemical compound which specifically binds to a mammalian NPFF receptor by a competitive binding method which comprises separately contacting cells containing DNA encoding and expressing on their cell surface the mammalian NPFF receptor, wherein such cells do not normally express the mammalian NPFF receptor, or a membrane preparation of such cells, with both the first chemical compound and a second chemical compound, under conditions suitable for binding of both compounds, and detecting specific binding of the first chemical compound to the mammalian NPFF receptor, a

decrease in the binding of the first chemical compound indicating that the first chemical compound binds to the mammalian NPFF receptor;

- (b) recovering the first compound free of any mammalian NPFF receptor; and
- (c) admixing a ~~pharmaceutically acceptable amount~~ ~~the compound with a pharmaceutically acceptable~~ carrier, thereby preparing the ~~pharmaceutical~~ composition;

wherein the mammalian NPFF receptor comprises an amino acid sequence which is the same as 1) the sequence of the human NPFF1 receptor encoded by plasmid pWE15-hNPFF1 (ATCC Accession No. 203183) or by plasmid pCDNA3.1-hNPFF1 (ATCC Accession No. 203605); 2) the sequence of the rat NPFF1 receptor encoded by plasmid pEXJ-rNPFF1 (ATCC Accession No. 203184); 3) the sequence of the human NPFF2 receptor encoded by plasmid pCDNA3.1-hNPFF2b (ATCC Accession No. 203255); or 4) the sequence shown in SEQ ID NO: 2, 4, 6 or 8.

--187: (Currently amended) A process for preparing a ~~pharmaceutical~~ composition which comprises:

- (a) identifying a compound that specifically binds to a mammalian NPFF receptor by a method which comprises contacting cells transfected with and expressing DNA encoding the mammalian NPFF receptor, wherein such cells do not normally

express the mammalian NPFF receptor, or a membrane preparation of such cells, with a first compound known to bind specifically to the mammalian NPFF receptor;

- (b) contacting the preparation of step (a) with a plurality of compounds not known to bind specifically to the mammalian NPFF receptor, under conditions permitting binding, and detecting specific binding of the first compound ~~known to bind to the mammalian NPFF receptor~~;
- (c) determining whether the binding of the first compound ~~known to bind to the mammalian NPFF receptor~~ is reduced in the presence of any compound within the plurality of compounds relative to the binding of the first compound in the absence of the plurality of compounds; and if so
- (d) separately determining the binding to the mammalian ~~mammalian~~ NPFF receptor of compounds included in the plurality of compounds so as to thereby identify a the compound included in the plurality of compounds which specifically binds;
- (e) recovering the compound which was included in the plurality of compounds free of any mammalian NPFF receptor; and
- (f) admixing a ~~pharmaceutically acceptable amount~~ ~~the compound with a pharmaceutically acceptable~~ carrier, thereby preparing the ~~pharmaceutical~~

composition;

wherein the mammalian NPFF receptor comprises an amino acid sequence which is the same as 1) the sequence of the human NPFF1 receptor encoded by plasmid pWE15-hNPFF1 (ATCC Accession No. 203183) or by plasmid pcDNA3.1-hNPFF1 (ATCC Accession No. 203605); 2) the sequence of the rat NPFF1 receptor encoded by plasmid pEXJ-rNPFF1 (ATCC Accession No. 203184); 3) the sequence of the human NPFF2 receptor encoded by plasmid pCDNA3.1-hNPFF2b (ATCC Accession No. 203255); or 4) the sequence shown in SEQ ID NO: 2, 4, 6 or 8.

--188: (Currently amended) A process for preparing a ~~pharmaceutical~~ composition which comprises:

- (a) identifying a chemical compound which specifically binds to and activates a mammalian NPFF receptor by a method which comprises contacting cells producing a second messenger response and expressing on their cell surface the mammalian NPFF receptor, wherein such cells do not normally express the mammalian NPFF receptor, with the chemical compound under conditions suitable for activation of the mammalian NPFF receptor, and measuring the second messenger response in the presence and in the absence of the chemical compound, a change in the second messenger response in the presence of the chemical compound indicating that the compound activates the mammalian NPFF receptor;

- (b) recovering the compound free of any mammalian NPFF receptor; and
- (c) admixing a ~~pharmaceutically acceptable amount~~ the ~~compound with a pharmaceutically acceptable~~ carrier, thereby preparing the ~~pharmaceutical~~ composition;

wherein the mammalian NPFF receptor comprises an amino acid sequence which is the same as 1) the sequence of the human NPFF1 receptor encoded by plasmid pWE15-hNPFF1 (ATCC Accession No. 203183) or by plasmid pcDNA3.1-hNPFF1 (ATCC Accession No. 203605); 2) the sequence of the rat NPFF1 receptor encoded by plasmid pEXJ-rNPFF1 (ATCC Accession No. 203184); 3) the sequence of the human NPFF2 receptor encoded by plasmid pCDNA3.1-hNPFF2b (ATCC Accession No. 203255); or 4) the sequence shown in SEQ ID NO: 2, 4, 6 or 8.

--189: (Currently amended) A process for preparing a ~~pharmaceutical~~ composition which comprises:

- (a) identifying a chemical compound which specifically binds to and inhibits activation of a mammalian NPFF receptor by a method which comprises separately contacting cells producing a second messenger response and expressing on their cell surface the mammalian NPFF receptor, wherein such cells do not normally express the mammalian NPFF receptor, with both the first chemical compound and a second chemical compound known to activate the NPFF receptor, and with only the second chemical

compound, under conditions suitable for activation of the mammalian ~~human~~ NPFF receptor, and measuring the second messenger response in the presence of only the second chemical compound and in the presence of both the second chemical compound and the first chemical compound, a smaller change in the second messenger response in the presence of both the first chemical compound and the second chemical compound than in the presence of only the second chemical compound indicating that the first chemical compound inhibits activation of the mammalian ~~human~~ NPFF receptor;

- (b) recovering the first compound free of any mammalian NPFF receptor; and
- (c) admixing a ~~pharmaceutically acceptable amount~~ the ~~compound with a pharmaceutically acceptable~~ carrier, thereby preparing the ~~pharmaceutical~~ composition;

wherein the mammalian NPFF receptor comprises an amino acid sequence which is the same as 1) the sequence of the human NPFF1 receptor encoded by plasmid pWE15-hNPFF1 (ATCC Accession No. 203183) or by plasmid pCDNA3.1-hNPFF1 (ATCC Accession No. 203605); 2) the sequence of the rat NPFF1 receptor encoded by plasmid pEXJ-rNPFF1 (ATCC Accession No. 203184); 3) the sequence of the human NPFF2 receptor encoded by plasmid pCDNA3.1-hNPFF2b (ATCC Accession No. 203255); or 4) the sequence shown in SEQ ID NO: 2, 4, 6 or 8.

composition which comprises:

- (a) identifying a compound which activates a mammalian NPFF receptor by a method which comprises contacting cells transfected with and expressing DNA encoding the mammalian NPFF receptor, wherein such cells do not normally express the mammalian NPFF receptor, with a plurality of compounds not known to activate the mammalian NPFF receptor;
- (b) determining whether the activity of the mammalian NPFF receptor is increased in the presence of such compounds; and if so
- (c) separately determining whether the activation of the mammalian NPFF receptor is increased by each compound included in the plurality of compounds, so as to thereby identify the compound that activates the mammalian NPFF receptor;
- (d) recovering the compound free of any mammalian NPFF receptor; and
- (e) ~~admixing a pharmaceutically acceptable amount the compound with a pharmaceutically acceptable carrier,~~ thereby preparing the ~~pharmaceutical~~ composition;

wherein the mammalian NPFF receptor comprises an amino acid sequence which is the same as 1) the sequence of the human NPFF1 receptor encoded by plasmid pWE15-hNPFF1 (ATCC Accession No. 203183) or by plasmid pcDNA3.1-hNPFF1 (ATCC Accession No. 203605); 2) the sequence of the rat NPFF1 receptor encoded by plasmid pEXJ-rNPFF1 (ATCC Accession No. 203184); 3) the

sequence of the human NPFF2 receptor encoded by plasmid pCDNA3.1-hNPFF2b (ATCC Accession No. 203255); or 4) the sequence shown in SEQ ID NO: 2, 4, 6 or 8.

--191: (Currently amended) A process for preparing a ~~pharmaceutical~~ composition which comprises:

- (a) identifying a compound that inhibits the activation of a mammalian NPFF receptor by a method which comprises contacting cells transfected with and expressing DNA encoding the mammalian NPFF receptor, wherein such cells do not normally express the mammalian NPFF receptor, with a plurality of compounds in the presence of a known mammalian NPFF receptor agonist, under conditions permitting activation of the mammalian NPFF receptor;
- (b) determining whether the activation of the mammalian NPFF receptor is reduced in the presence of such plurality of compounds, relative to the activation of the mammalian NPFF receptor in the absence of the plurality of compounds; and if so
- (c) separately determining the inhibition of activation of the mammalian NPFF receptor for each compound included in the plurality of compounds is increased by each compound included in the plurality of compounds, so as to thereby identify a ~~the~~ compound that inhibits the activation the mammalian NPFF receptor;
- (d) recovering the compound free of any mammalian NPFF receptor; and

Applicants: Christophe P.G. Gerald, et al.
Serial No.: 09/866,248
Filed: May 25, 2001
Page 14

- (e) ~~admixing a pharmaceutically acceptable amount the compound with a pharmaceutically acceptable carrier,~~
thereby preparing the pharmaceutical composition;

wherein the mammalian NPFF receptor comprises an amino acid sequence which is the same as 1) the sequence of the human NPFF1 receptor encoded by plasmid pWE15-hNPFF1 (ATCC Accession No. 203183) or by plasmid pCDNA3.1-hNPFF1 (ATCC Accession No. 203605); 2) the sequence of the rat NPFF1 receptor encoded by plasmid pEXJ-rNPFF1 (ATCC Accession No. 203184); 3) the sequence of the human NPFF2 receptor encoded by plasmid pCDNA3.1-hNPFF2b (ATCC Accession No. 203255); or 4) the sequence shown in SEQ ID NO: 2, 4, 6 or 8.